K-13

Differential profiling of breast cancer plasma proteome by isoepitope-coded affinity tagging method

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Breast cancer rate is increasing in Korea. The problem is not only increase of number but the increase rate is so fast. The advent of proteomics provides the hope of discovering novel biological markers that can be used for early detection, disease diagnosis and prediction of response to therapy. We examined breast cancer plasma proteome for discovery of novel biomarker candidates, using the cleavable isoepitope-coded affinity tag (cICAT) labeling strategy and LC-MS/MS. Plasmas from 7 breast cancer patients and 6 healthy women were gathered and pooled separately. After depletion of six abundant plasma proteins using multiple affinity removal system, the plasma samples were labeled with cICAT and analyzed by LC-MS/MS. A total of 114 proteins were identified and quantified, with 24 proteins exhibiting statistically significant abundance change. Among these, 12 were up-regulated by more than 1.5-fold and 12 were down-regulated by the same fold in breast cancer plasma. The proteins could be categorized by their biological functions; the main groups correspond to immune response, metabolism, cell adhesion and cytoskeleton, proteolysis, coagulation, transport and signaling. Our results provide candidates for potential biomarkers that are useful in diagnosis of breast cancer.

K-14

Differential proteome analysis of human colorectal cancer by two-dimensional difference in gel electrophoresis and tandem mass spectrometry

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Colorectal cancer (CRC) is one of the most common malignant tumors in the world. It has a high 3-year mortality rate and a low early stage diagnosis rate. It still has a poor prognosis, for 50% of the cases are reported to be incurable at the time of diagnosis. In this study, we used two-dimensional difference in gel electrophoresis (2D-DIGE) and tandem mass spectrometry to investigate differentially expressed proteins in CRC. A total of 6 cancerous tissues and 6 matched non-tumorous surrounding mucosa areas were obtained from 6 volunteering patients. Matrix-assisted laser desorption/ionization-time of flight (MALDI-TOF) and tandem (TOF/TOF) mass spectrometry provided sensitive and accurate mass spectral data for database interrogation, resulting in the identification of 46 different proteins that showed changes in the level of expression between tumor and nontumor tissues. 28 proteins were up-regulated and 18 were down-regulated in CRC. The proteins could be categorized according to their functions into a variety of pathways including metabolism, transport and signaling, cell cycle regulation, cytoskeleton, and stress. Identification of potential biomarkers provides further useful insights into the pathogenesis of CRC.

K-15

Epitope analysis of the major allergenic protein fag e 1 from autogamous common buckwheat

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In the present study, we focused on the molecular characterization of Fag e 1 for the purpose of developing hypoallergenic buckwheat. Fag e 1 cDNA was isolated from autogamous common buckwheat and its antigenicity confirmed by immunoblotting using recombinant protein expressed in E. coli. The derived amino acid sequence from Fag e 1 cDNA has been used to construct the synthetic peptides, and we have identified the major IgE binding epitopes in this allergenic protein. We isolated the respective cDNA, coding for a 22 kDa protein, from a recently developed autogamous strain of common buckwheat and confirmed its immunoglobulin E (IgE)-binding activity using recombinant Fag e 1 and sera of allergic patients. The derived amino acid sequence from Fag e 1 cDNA was used to synthesize an overlapping peptide library on nitrocellulose membranes for the determination of the Fag e 1 epitopes. We identified eight epitopes and the critical amino acids for IgE-binding within the epitopes. This epitope analysis of a major allergenic protein of buckwheat should help therapeutic efforts and aid in the development of hypoallergenic buckwheat.

K-16

Genomic and proteomic analysis of hepatocellular carcinoma-related biomarkers in the liver of HBx Tg mice

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Hepatitis B virus (HBV) has been clearly recognized as an etiological factor for hepatocellular carcinoma (HCC). HBV encodes the potentially onconeogenic HBx protein. However, little was known for the mechanism underlying HBx-mediated oncoenergetic. Although there have been many attempts to investigate the role of HBx, however, in vivo the role of HBx is not fully understood during hepatocellular carcinogenesis. Therefore, we aimed to elucidate the molecular mechanism of HCC caused by HBx and to discover the biomarker related to HCC by HBx. To find differentially expressed genes and proteins during hepatocellular carcinoma, we employed oligo DNA microarray and 2DE proteomics with 3, 9 and 13 month-old HBx Tg mice. To investigate the transcriptional patterns occurring during carcinogenesis of HCC, we identified nine clusters representing the different gene expression patterns across the time course using hierarchical clustering. We found total 37 differentially expressed proteins. Some key proteins involved in iron metabolism, anti-oxidant enzyme, amino acid metabolism and cell proliferation. Our results may provide useful information for the understanding of molecular pathogenesis of HCC driven by HBx.